ABSTRACT

As one of the most useful tools to influence gut microbiota and body health, probiotics have been studied for over a century in the aspect of different diseases. Many animals experiments and randomized clinical trials(RCTs) are conducted to demonstrate their therapeutical effect. This systematic review discussed the effect of probiotics on three aspects of human health, obesity, type II diabetes(T2D) and digestive health. In this review, more than 20 randomized clinical trials are included and involved in a meta-analysis using STATA 14.0 software. The outcome suggested that intervention of probiotics has beneficial effects on improving obesity and type II diabetes, but shows little effect on digestive health. However, due to the insufficient data collected in this meta-analysis, no definitive conclusion can be drawn and more RCTs are necessarily needed to investigate the effect of probiotics on human health in greater detail.

BIOGRAPHICAL SKETCH

Yalin Xiao finished her undergraduate study at China Agricultural University in 2018 majoring Food Science and Engineering. After graduation, she continued her study in food science at Cornell University as an M.P.S. student in Professor Rui Hai Liu's lab.

Thanks to Professor Liu's great help and instruction.

Thanks to all my dear friends who support me no matter what.

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INTRODUCTION

1.1 Probiotics

According to the Food and Agriculture Organization (FAO)/World Health Organization (WHO), probiotics are defined as "live microorganisms which, when administered in adequate amounts, confer a health benefit on the host". Guidelines are also launched regarding the evaluation of probiotics: for an adequate amount of health benefits, a dose of 5 billion colony forming units (cfu) has been recommended for at least 5 days ($5 \cdot 10^9$CFU/day)[1]. To identify the benefits that probiotics can provide in human health, many researches have been conducted in recent decades. Early researches of probiotics were mostly concentrated on the nutritive functions as a form of food or dietary supplement[2]. Recently, the trend in research interest of probiotics has shifted to the therapeutic and medicinal purposes of probiotics, as a scientific evolution in gut microbiome and probiotics research has occurred[3]. There are a large number of experiments or clinical trials to figure out whether probiotics have effects on some certain kinds of diseases or metabolism abnormalities. A general conclusion is that probiotics can provide a wide range of benefits to human health, especially in the aspect of gut-related cases.

The most studied and used probiotics in recent years are lactobacilli and Bifidobacterium. In addition to these two, Lactobacillus acidophilus, Lactobacillus salivarius, Lactobacillus rhamnosus, Lactobacillus Plantarum, Lactobacillus bulgaricus, Lactobacillus casei, Lactobacillus sporogenes, Bifidobacterium longum, Bifidobacterium infantis, Bifidobacterium bifidum, Streptococcus thermophilus and Homeostatic Soil Organisms (HSO's) are also commonly used in probiotics formulations[1]. A good probiotic needs to meet several criteria as follows: 1) it is capable of adhering to cells; 2) It can exclude or reduce adherence of pathogens; 3) It is able to persist, multiply and produce acids, hydrogen peroxide and bacteriocins that are antagonistic to pathogen growth; 4) It is safe, noninvasive, non-pathogenic and noncarcinogenic； 5) it can co-aggregate to produce a balanced microbiota[4].

As a result of these researches, many kinds of probiotics products are developed. Yogurt and other dairy food rich in probiotics are well known to improve gut health, by strengthening the intestinal barrier function and stimulating the immune system[5]. “Probiotic dietary supplements” is regarded as the second largest probiotics market, following “probiotic food and beverage”[6]. Certain species of probiotics are made into tablets that can be taken by patients of certain kinds of diseases to help them improve body health.

The aim of this review is to summarize three aspects that probiotics can be beneficial to human health, which are obesity, type II diabetes, and digestive health, by looking into related experiments and clinical researches.

1.2 Obesity

Nowadays, obesity has become a pandemic issue; more than 1.9 billion people are overweight. The most common cause of obesity is the imbalance between energy intake and energy expenditure. Genetic and environmental factors, including the fat mass- and obesity- associated gene FTO, low level of physical activity, high-fat diet and so on, are also important causes of obesity. Obese patients usually face greater risks of cardiovascular diseases, type 2 diabetes (T2D), dyslipidemia, hepatobiliary disease (non-alcoholic fatty liver disease, gallbladder dyskinesia, cholelithiasis) as well as lung, breast, uterine and ovarian cancer. In previous studies, it was mainly focused on environmental and dietary factor which could influence energy balance. With the understanding of obesity and human health developing, gut microbiota is being taken into consideration due to their effect on energy homeostasis, inflammation and so on, suggesting a new method against obesity.

A large amount of scientific evidence indicates that the composition of gut microbiota is associated with obesity. In both animal and human studies, a higher abundance of Firmicutes, especially Lactobacilli is observed, with a lower proportion of the Bacteroidetes in obesity. Due to the fact that gut microbiota composition varies among everyone, the general situation is quite changeable in obese people[5].

N. Kobyliak et al., demonstrated that probiotics mixture and with nutraceutical have led to a significantly lower prevalence of obesity, reduction of insulin resistance, total and visceral adipose tissue (VAT) weight in the rat monosodium glutamate (MSG) obesity model. Supplementation of probiotics with omega-3 may have the most beneficial anti-obesity properties in their experiment[7]. However, not all the genera of probiotics are beneficial for losing weight. Taking the genera of Lactobacillus as an example, the effects of Lactobacillus vary according to the species: some species were linked to weight gain, while others were associated with weight loss[8]. Some clinical trials indicate that administration of Lactobacillus reuteri, Lactobacillus sakei, Lactobacillus acidophilus and L. casei was associated with weight gain in human, whereas consumption of Lactobacillus gasseri, Lactobacillus amylovorus and L. plantarum was associated with weight loss in obese humans, body fat loss in overweight people and in healthy individuals, respectively[5].

1.3 Type II Diabetes

Type II diabetes mellitus is a metabolic disease which begins with increased insulin resistance (IR); patients of Type II diabetes mellitus(T2DM) cannot have their cells respond to insulin properly, and develop a high blood sugar level in the long term[9]. Family history of diabetes, overweight, and obesity, unhealthy diet, physical inactivity and smoking are the strongest risk factors for Type II Diabetes (T2D). It was estimated by WHO that 422 million adults aged over 18 years were living with diabetes in 2014 around the whole world (International diabetes federation. IDF diabetes atlas. 7th edition Diabetes Atlas; 2015. http://www.diabetesatlas.org/.). This pandemic health issue has been studied for decades, from aspects including insulin and enzymes, and many therapeutical methods have been put into use. It caught people's attention that compared to non-diabetic patients, T2D patients have altered gut microbiota composition, mostly with a lower relative abundance of Firmicutes and a higher proportion of Bacteroidetes and Proteobacteria[7]. It is revealed by some animal studies that altered microbiota may contribute to the pathogenesis of insulin resistance and thereby T2D by several mechanisms. Due to the alteration of gut microbiota given by probiotics, people started to study the relationship between probiotics and T2D.

Some researches demonstrate that probiotics can help to manage the gut microbiota environment and increase the production of short chain fatty acids (SCFAs), like acetate and butyrate, which can decrease insulin resistance and promote pancreatic cells proliferation and development, by activating certain receptors. Both regulated gut microbiota and SCFAs can stimulate the secretion of GLP-1 and GLP-2, thus increasing insulin and adiponectin expression, and might contribute to the enhanced insulin sensitivity, as well as decreasing low-grade inflammation associated with T2D.

1.4 Digestive Health

Digestive health is a general concept including a wide range of diseases, such as ulcerative colitis (UC), Crohn's disease (CD), diarrhea, constipation and so on. Most digestive diseases are claimed to be associated with gastrointestinal tract. For example, UC and CD are two representative causes of chronic inflammation of gastrointestinal tract, leading to diarrhea, abdominal pain and other symptoms. Newborn babies have sterile gastrointestinal tract; bacteria ingested during the birth process will colonize the gastrointestinal tract. While babies grow, their intestinal flora becomes similar to their mothers' intestinal flora gradually[10]. Microbiota in intestines is responsible for functioning the immune system properly. Considering the fact that probiotics have great influence on gut microbiota and intestinal environment, researchers started to study whether probiotics can be used to treat digestive health problems, including constipation and diarrhea.

For patients who go through radiotherapy at abdominal or pelvic cavity, radiation-induced enteritis is one of the most common complications. Seher Demirer, et al., have used lactobacillus bulgaricus strain isolated from yogurt along with radiation-therapy Wistar rats models, demonstrated that probiotics might have some beneficial effects against radiation-induced intestinal injury by accelerating healing ability and preventing bacterial translocation and diarrhea[11].

Despite that several studies claim probiotics could be beneficial to gastrointestinal health, many research hold the point that the evidence is not enough to prove it. American College of Gastroenterology has conducted a meta-analysis of 53 randomized, controlled trials involving 5545 patients with irritable bowel syndrome(IBS)[12]. The results of the analysis suggest that considered all together, probiotics were statistically superior to placebo with a relative risk of persistence of symptoms, but not significantly. After analyzing specific strands or combinations of probiotics, such as Lactobacillus Plantarum, Escherichia coli, Streptococcus faecium, and Bifidobacterium, researchers claimed that total adverse events were not statistically higher with probiotics comparing to placebos. The final statement of ACG was that probiotics may improve global symptoms, as well as bloating and flatulence in IBS patients, but the scientific evidence was not strong enough to confirm it.

In a word, probiotics are a potential therapy for digestive health problems, due to its effect on gut microbiota composition and intestinal mucosa. While we have just started to dive into this research area, more studies and trials are needed to be conducted to provide deeper views.

METHODS AND MATERIALS

2.1 Searching strategy and selection of RCTs for meta-analysis

Databases used in this review includes PubMed, Web of Science, the Cornell Library, ClinicalTrials.gov.

Literature was searched under such criteria[13]:

-Types of study: randomized clinical trials.

-Types of intervention: probiotics supplementation without restriction on dosage, strains, methods of administration.

-Duration of study: longer than 3 weeks.

-Types of control: placebo.

2.2 Data collection process

Based on literature search in the database, the following data were collected:

-Study identification including author name and year of publication.

-Study design information including country, duration of intervention, sample size and characters in each group, probiotics strains and dosage used in the intervention.

-Primary outcomes and secondary outcomes including detailed data of related factors.

Literature without sufficient data was excluded in meta-analysis.

Studies with sufficient data were include in forest plots made via STATA 14.0 software. Factors collected were as following:

-Obesity: Body Mass Index(BMI), waist circumference(WC), fat mass.

-Type II diabetes: Homeostasis Model Assessment-Insulin Resistance(HOMA-IR), insulin, systolic blood pressure(SBP) and diastolic blood pressure(DBP).

-Digestive health: stool consistency, frequency of defecation, abdominal pain, colonic transit time(CTT).

RESULTS

3.1 Summary of Study

Among literature identified, 8 RCTs about obesity, 6 RCTs about type II diabetes, and 7 RCTs about digestive health in total were included and the main design information was summarized in Table 1, 2, 3.

3.2 Effect of probiotics on obesity

7 RCTs in total have reported the Body Mass Index (BMI) in their results. Administration of probiotics has led to a significant decrease in BMI (-0.44, 95% CI: -0.67, -0.20) (Fig. 1). Waist circumference was also reported in 7 RCTs, which is proved to be decreased significantly after the intervention of probiotics (-1.57 cm, 95% CI: -2.20, -0.94) (Fig. 2). A decrease was noted in the effect of probiotics on body fat mass (-1.23, 95% CI: -2.75, 0.30), however, it is not significant (Fig. 3).

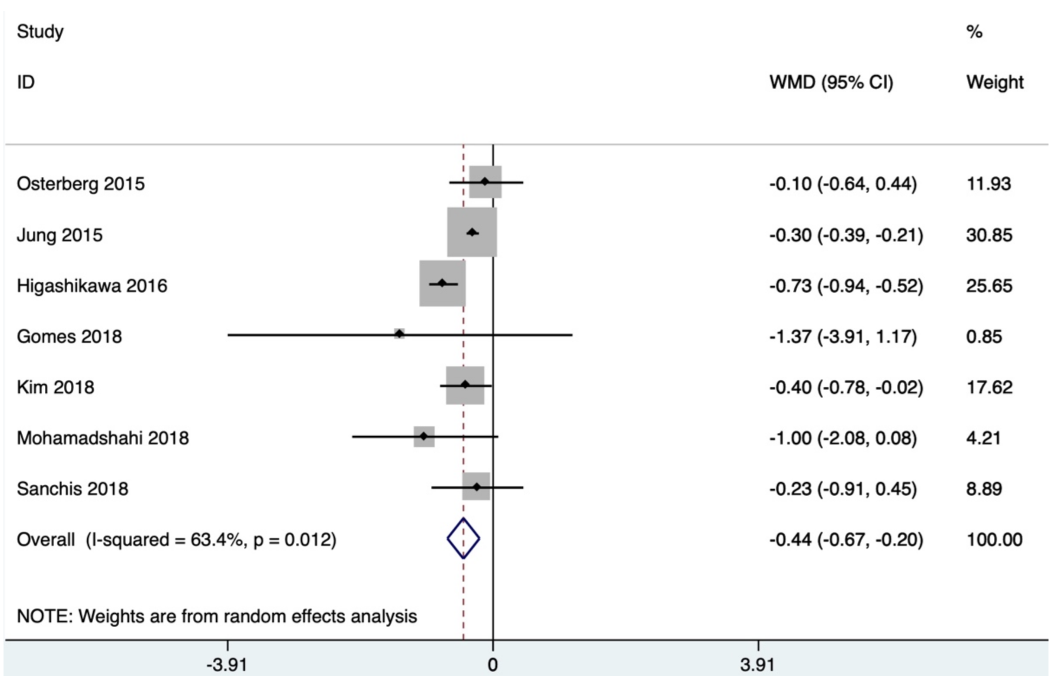

Fig. 1. Forest plot of BMI that compares between probiotics intervention group and placebo group.

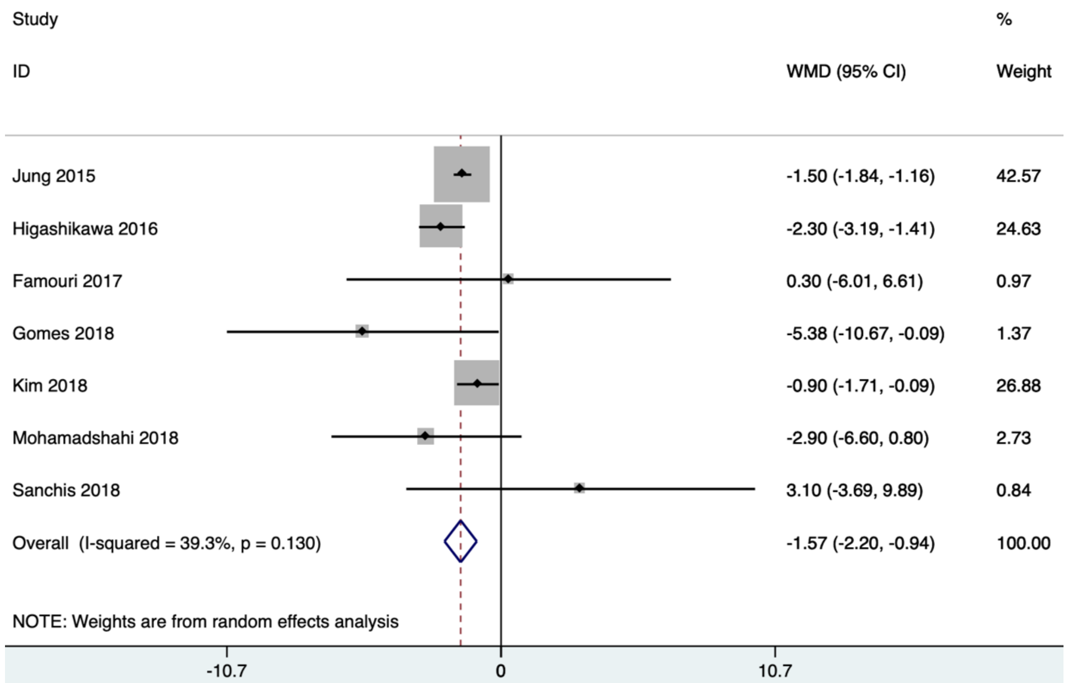

Fig. 2. Forest plot of waist circumference that compares between the probiotics intervention group and placebo group.

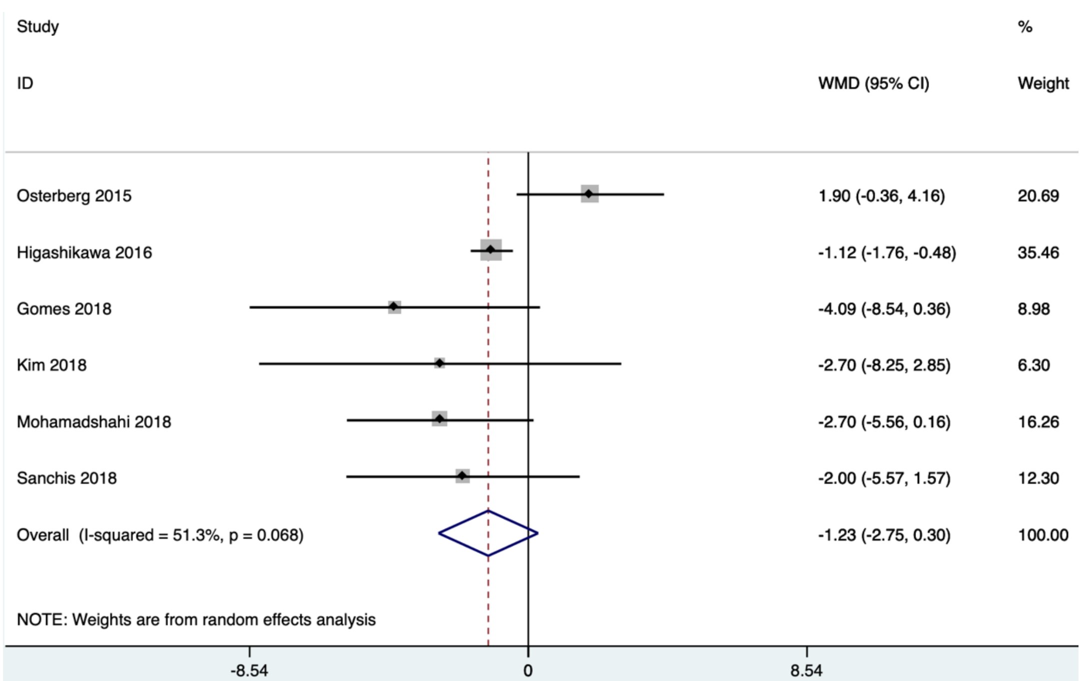

Fig. 3. Forest plot of Fat Mass that compares between the probiotics intervention group and placebo group.

3.3 Effect of probiotics on T2D

As one of the most important factor in T2D, HOMA-IR (Homeostasis Model Assessment-Insulin Resistance) showed a significant decrease (-1.25, 95%CI: -2.48, -0.23) in probiotics intervention group overall in Fig. 4, despite that one study in 2017 held the opposite outcome. Blood insulin level was also greatly decreased by probiotics (-2.88, 95%CI: -5.68, -0.09) (Fig.5). However, systolic blood pressure (SBP) and diastolic blood pressure (DBP) failed to show any significant change due to the intake of probiotics (Fig. 6 and Fig. 7).

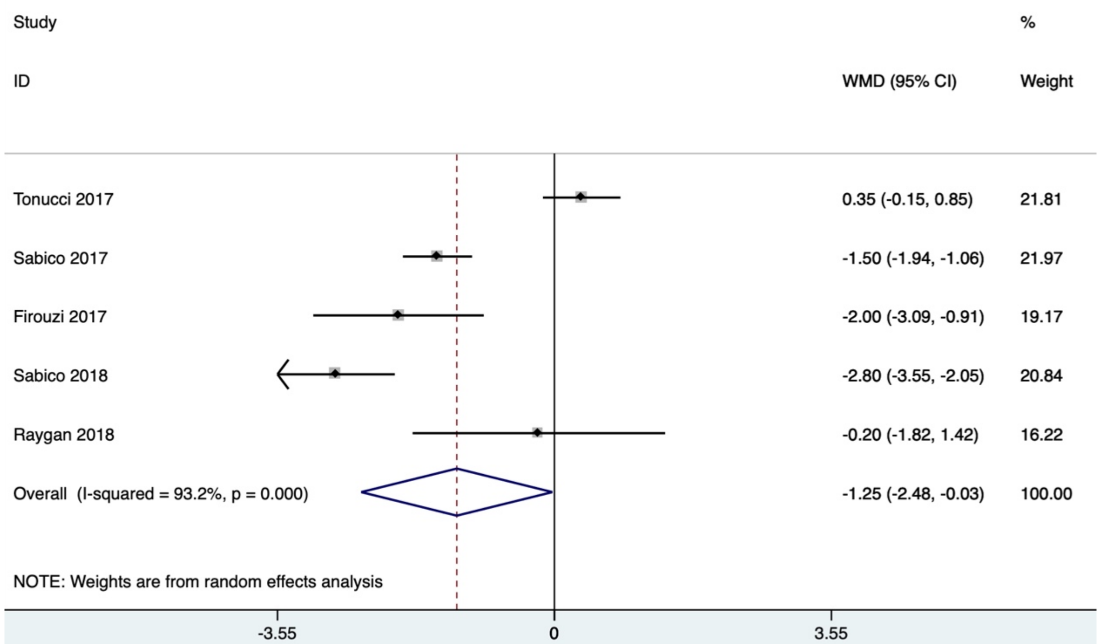

Fig. 4. Forest plot of HOMA-IR that compares between the probiotics intervention group and placebo group.

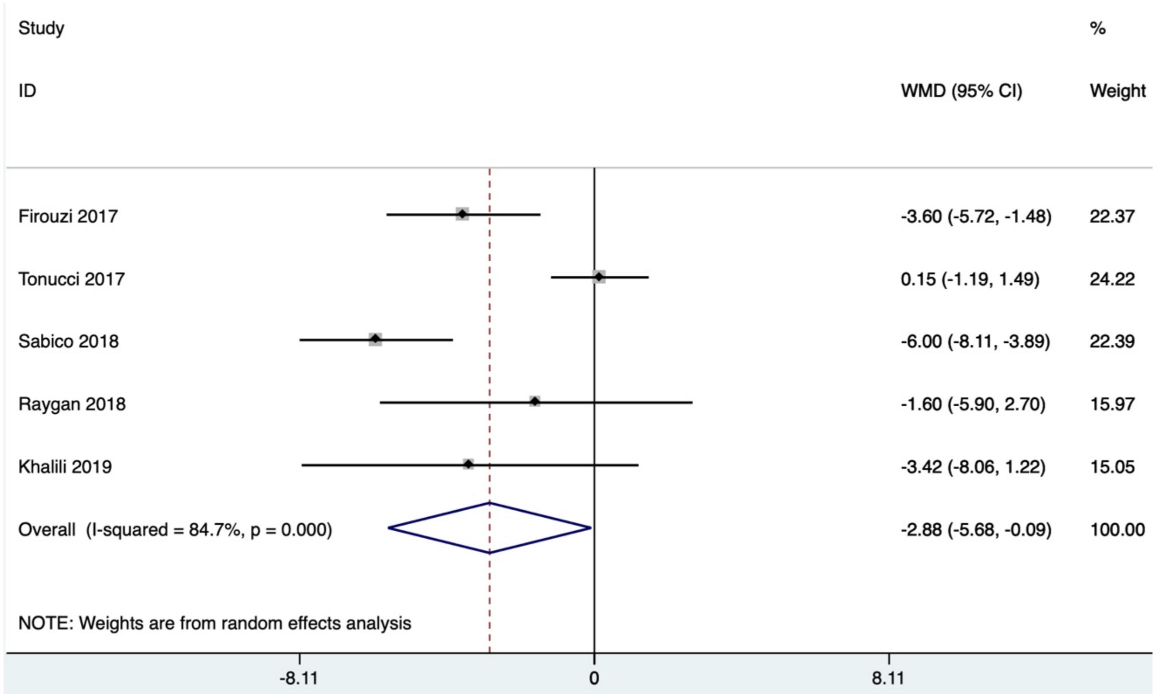

Fig. 5. Forest plot of insulin that compares between the probiotics intervention group and placebo group.

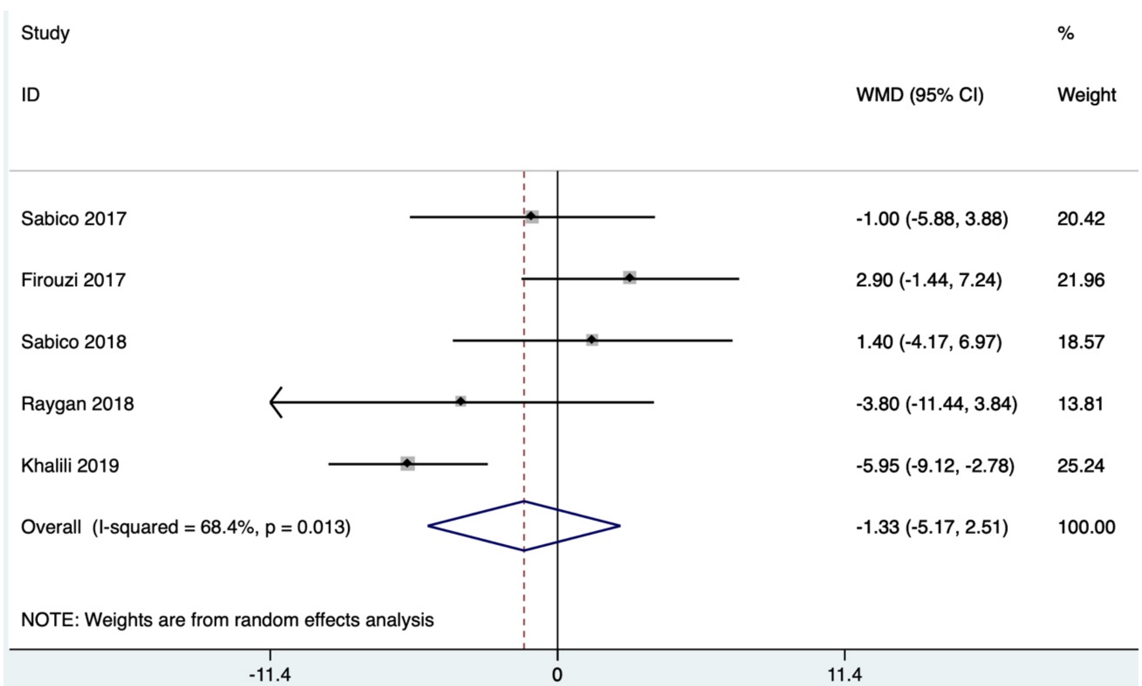

Fig. 6. Forest plot of systolic blood pressure that compares between the probiotics intervention group and placebo group.

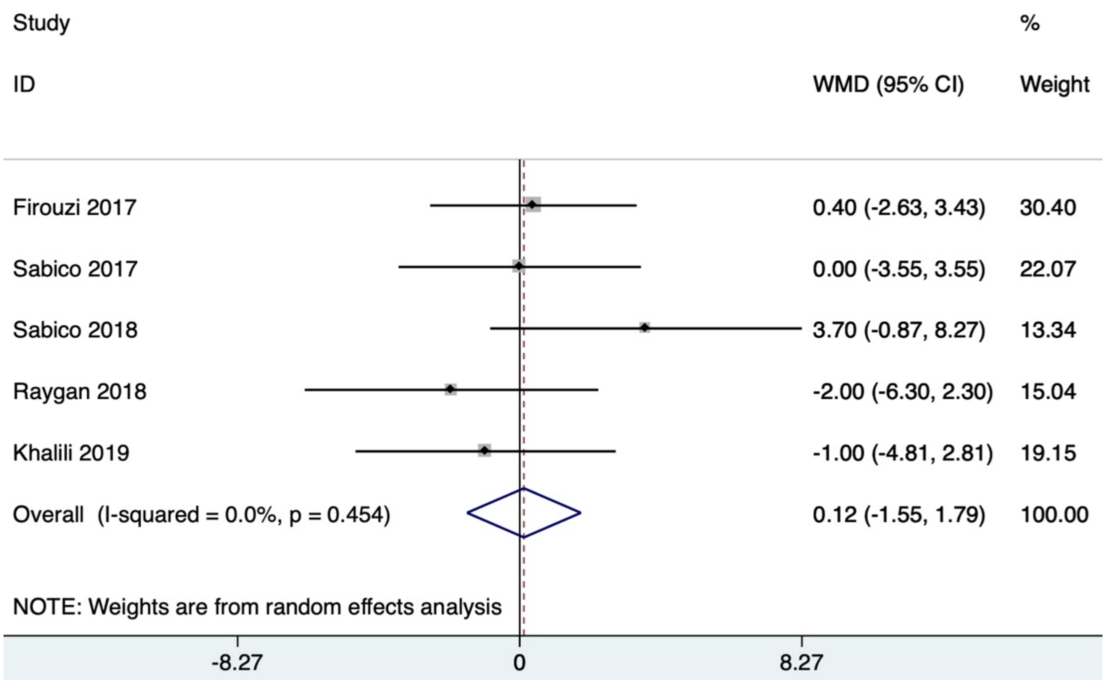

Fig. 7. Forest plot of diastolic blood pressure that compares between the probiotics intervention group and placebo group.

3.4 Effect of probiotics on digestive health

4 RCTs have reported stool consistency measured by the Bristol Stool Scale. After the administration of probiotics, stool consistency was significantly improved by 0.56 (95% CI: 0.06, 1.03) (Fig. 8). Frequency of defecation was also improved overall by 0.32 (95% CI: -0.32, 0.96), but not significantly enough (Fig. 9). No great impact was detected in aspect of abdominal pain and bloating (-0.04, 95% CI: -0.19, 0.10; 0.59, 95% CI: -0.40, 1.58, respectively) (Fig. 10 and Fig. 11). Colonic transit time (CTT) was decreased by probiotics by 2.49 (95% CI: -24.18, 19.21), but a great inconsistency was observed among these trials (Fig. 12).

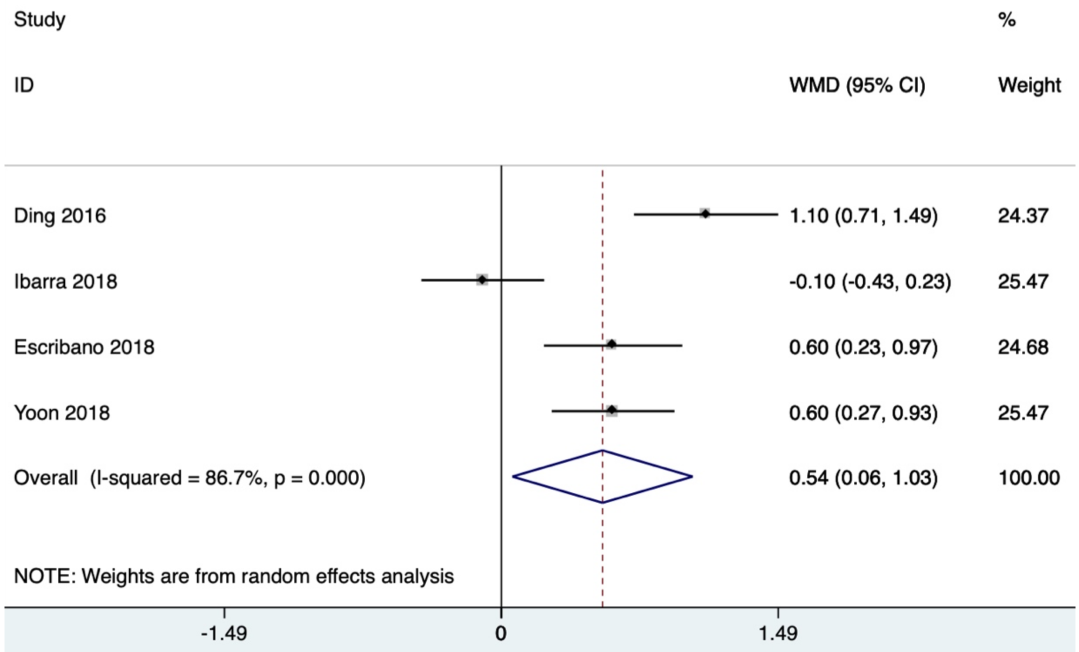

Fig. 8. Forest plot of stool consistency that compares between the probiotics intervention group and placebo group.

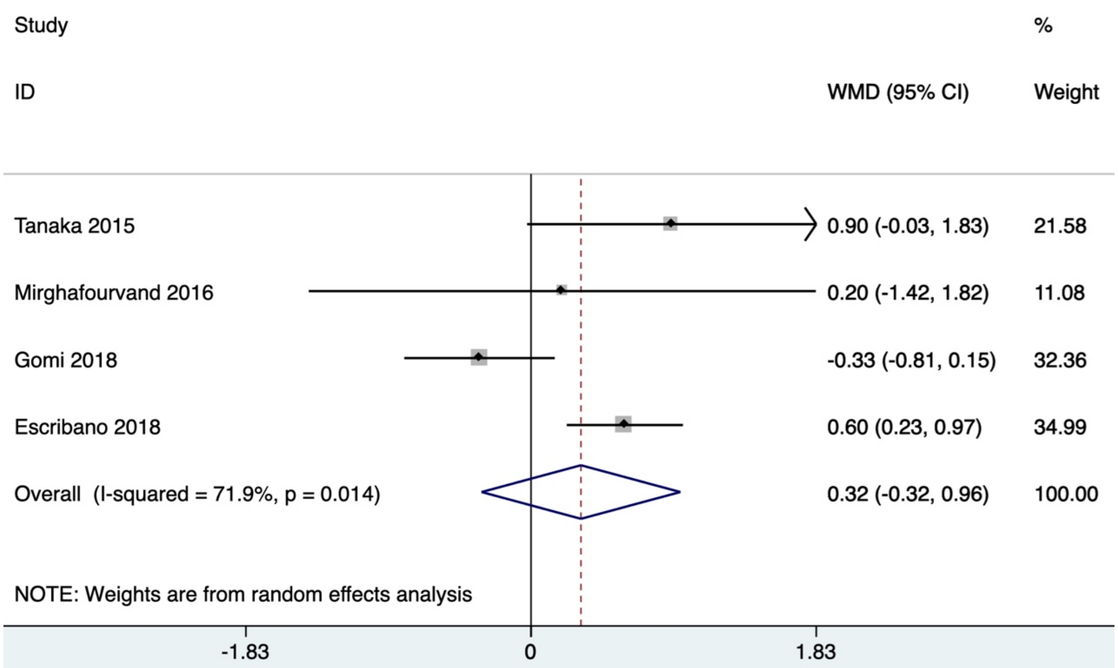

Fig. 9. Forest plot of the frequency of defecation that compares between the probiotics intervention group and placebo group.

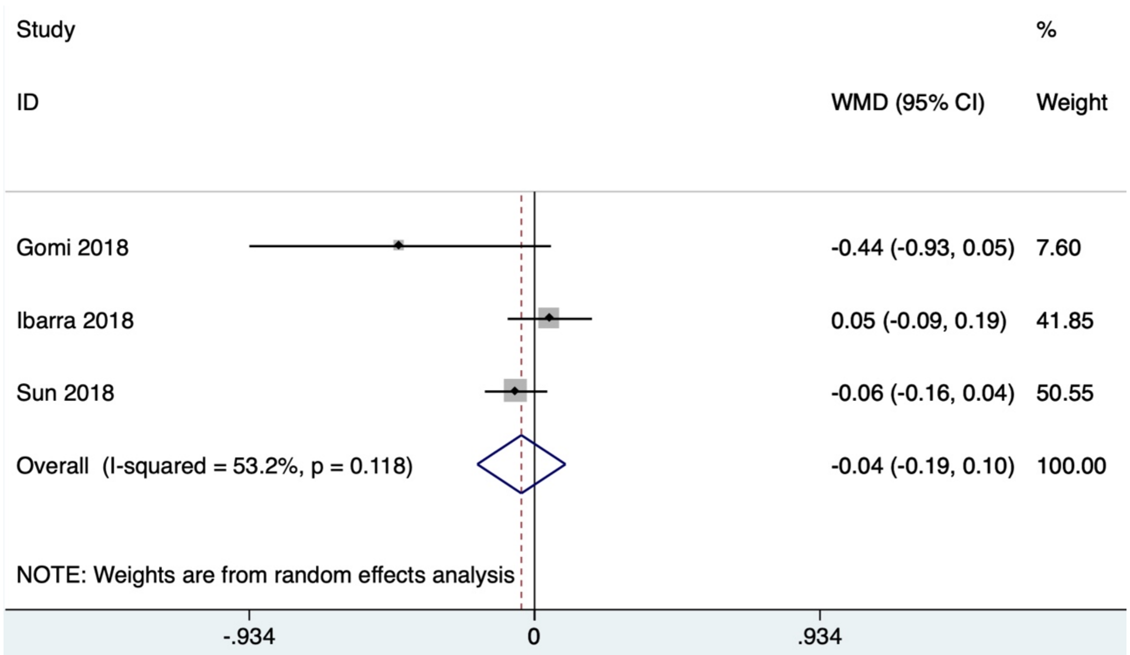

Fig. 10. Forest plot of abdominal pain that compares between the probiotics intervention group and placebo group.

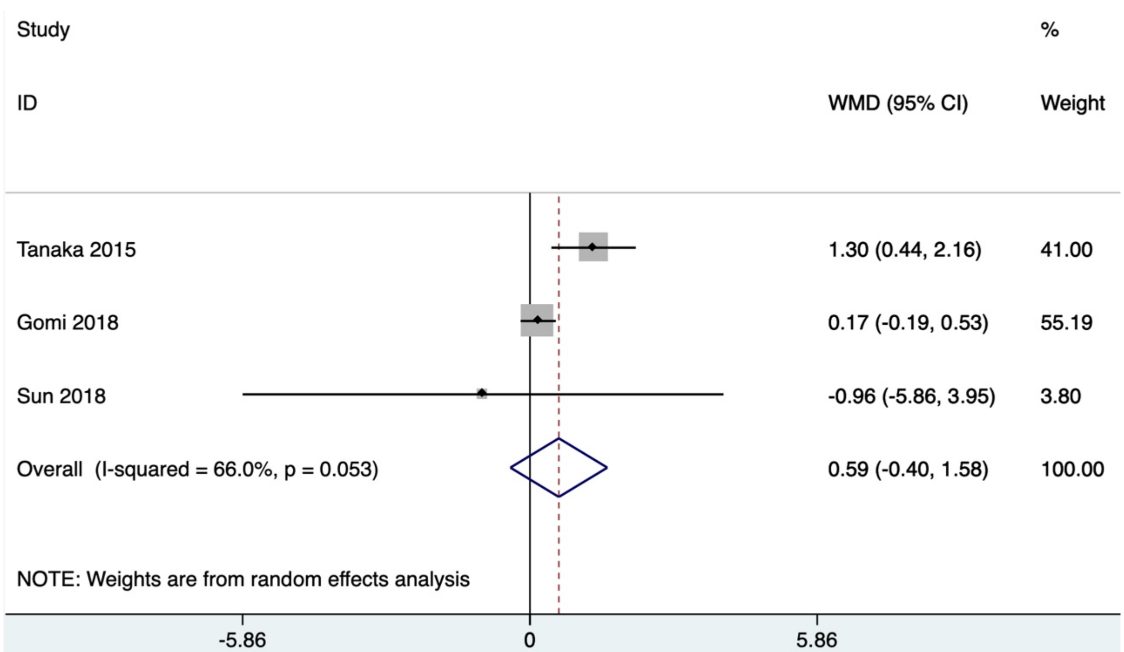

Fig. 11. Forest plot of bloating that compares between the probiotics intervention group and placebo group.

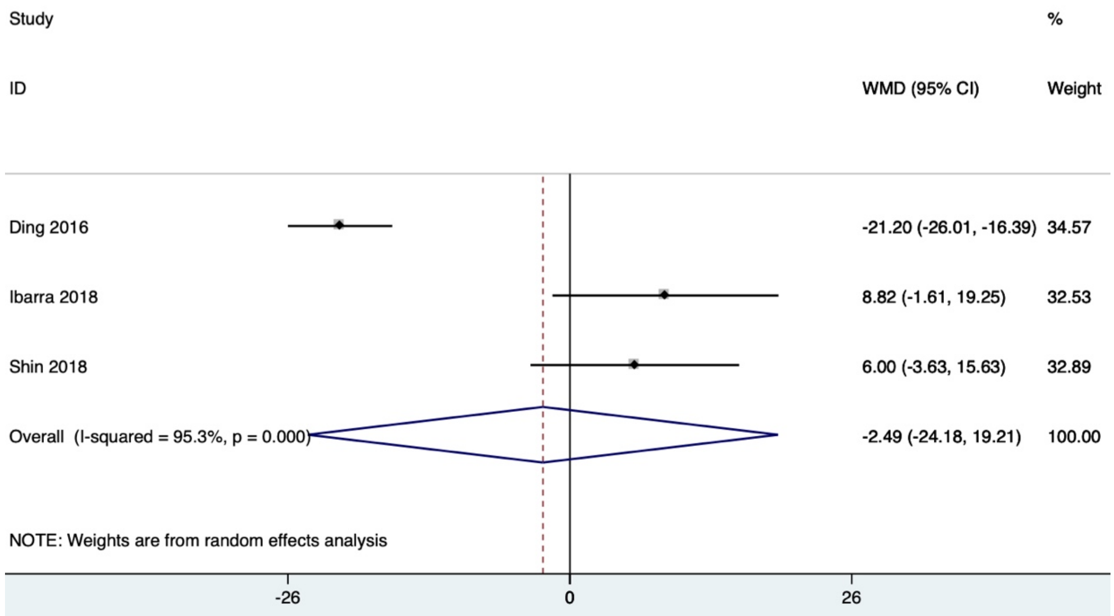

Fig. 12. Forest plot of colonic transit time that compares between the probiotics intervention group and placebo group.

Table 1 Summary of RCTs about effect of probiotics on obesity.

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Osterberg et al 2015[14]	the United States	Randomized, double-blind placebo-controlled trial, 4 weeks	Healthy non-obese people (BMI <30) Age: 18-30 y Male: 100%	VSL#3® (Streptococcus thermophilus DSM24731, L. acidophilus DSM24735, L. delbrueckii ssp. bulgaricusDSM24734, L. paracasei DSM24733, L. plantarum DSM24730, B. longum DSM24736, B. infantis DSM24737, and B. breve DSM24732)	Total: 20 Placebo group (high-fat, hypercaloric diet + placebo): n=11 Intervention group(high-fat, hypercaloric diet + probiotics intervention): n=9	Two sachets of VSL#3 (450 billion bacteria per sachet)/day	Placebo+ high-fat, hypercaloric diet	Intervention group: body mass and fat mass increased less comparing with placebo group but no significant changes in insulin sensitivity or in vitro skeletal muscle pyruvate and fat oxidation was detected.
Jung et al 2015[15]	Korea	Double-blind, placebo-controlled, randomized trial, 12 weeks	Nondiabetic overweight subjects Male: 34 (35.79%) Female: 61 (64.21%)	L. curvatus HY7601 plantarum KY1032	Total: 95 Placebo group: n=46 Intervention group: n=49	L. curvatus HY7601 at 2.5 × 10^9 cfu/day and L. plantarum KY1032 at 2.5 × 10^9 cfu/day	Placebo	Intervention group: reduced body weight (−0.65 ± 0.23 kg), body fat percentage (−0.57 ± 0.19%), body fat mass (−616 ± 161 g) , and L1 subcutaneous fat area (−2.68 ± 1.31 cm^2).
Higashikawa et al (2016)[16]	Japan	Randomized, double-blind, placebo-controlled clinical trial, 12 weeks	Age: 20–70 y Male: 23 (37%) Female: 39 (63%)	LP28: plant-derived lactic acid bacterium (Pediococcus pentosaceus)	Total: 62 Placebo group: n=20 Intervention group I (living LP28): n=21 Intervention group II (heat-killed LP28) : n=21	10^{11} CFU/day	Placebo	Intervention group II (heat-killed LP28): reduced BMI (0.45 kg/m2, 95% CI (0.04, 0.86), p=0.035), body fat percentage (1.11%, (0.39, 1.82), p=0.002), body fat mass (1.17 kg (0.43, 1.92), p=0.004) and waist circumference (2.84 cm (0.74, 4.93), p=0.009).
Famouri et al 2017[17]	Iran	Randomized, triple-blind, placebo-controlled clinical trial, 12 weeks	Obese children with sonographic NAFLD Age: 20-18 y Male: 32 (50%) Female: 32 (50%)	Lactobacillus acidophilus ATCC B3208, Bifidobacterium lactis DSMZ 32269, Bifidobacterium bifidum ATCC SD6576, Lactobacillus rhamnosus DSMZ 21690.	Total: 64 Placebo group: n=32 Intervention group: n=32	Lactobacillus acidophilus ATCC B3208, 3 × 10^9 cfu; Bifidobacterium lactis DSMZ 32269, 6 × 10^9 cfu; Bifidobacterium bifidum ATCC SD6576, 2 × 10^9 cfu; Lactobacillus rhamnosus DSMZ 21690, 2 × 10^9 cfu	Placebo	Intervention group: decreased mean cholesterol, low-density lipoprotein-C, and triglycerides as well as waist circumference; no significant change in weight, body mass index, and body mass index z score was detected.

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Table 1 (continued)

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Gomes et al (2018)[18]	Brazil	Randomized, double-blind, placebo-controlled, two-arm, parallel-group study, 8 weeks	Women with excess weight or obesity Age: 20-59 y	Lactoba- cillus acidophilus LA-14, Lactobacillus casei LC-11, Lactococcus lactis LL-23, Bifidobacterium bifidum BB-06, and Bifidobacterium lactis BL-4 (DaniscoVR®)	Total: 43 Placebo group (dietary intervention): 22 Intervention group(dietary + probiotics intervention): 21	2×10^{10} CFU/day	Placebo+ dietary intervention	Intervention group(dietary+probiotics intervention): reduced waist circumference (p=0.03), waist-height ratio (p=0.02), conicity index (p=0.03), plasma polyunsaturated fatty acids (p=0.04); increased activity of glutathione peroxidase ($p < 0.01$).
Mohammadi-Sartang et al 2018[19]	Iran	Randomized double-blinded controlled trial, 10 weeks	Obese adults Male: 34 (39.1%) Female: 53 (60.9%)	Bifidobacterium lactis Bb- 12	Total: 87 Placebo group: 43 Intervention group: 44	2×10^{7} cfu/g	Placebo	Intervention group: reduced body fat mass (p=0.023), body fat percentage (p=0.028), waist circumference (p=0.002), HOMA-IR (p=0.025), triglyceride concentration (p=0.003).
Sanchis et al 2018[20]	Spain	Randomized, double-blind, placebo-controlled clinical trial, 13 weeks	Obese children Age: 10-15 y Male: 24 (50%) Female: 24 (50%)	B. pseudocatenulatum CECT 7765	Total: 48 Placebo group: n=25 Intervention group: n=23	$1 \times 10^{9} \sim 1 \times 10^{10}$ colony forming units (CFU)	Placebo	Intervention group: reduced circulating high-sensitive C-reactive protein (p=0.026) and monocyte chemoattractant protein-1 (P=0.032); increased high-density lipoprotein cholesterol (Pp=0.035) and omentin-1 (P=0.023)
Kim et al (2018)[21]	Korea	Randomized, double-Blind, placebo-controlled trial, 12 weeks	Obese adults Age: 20–75 y Female: 63 (70%) Male: 27 (30%)	Lactobacillus gasseri BNR17	Total: 90 Placebo group: 30 Intervention group I (high dose): 30 Intervention group II (low dose): 30	Intervention group I (high dose): 1×10^{10} CFU/day Intervention group II (low dose): 1×10^{9} CFU/day	Placebo	Intervention group I (high dose): Reduced waist circumferences significantly (P=0.012); decreased visceral adipose tissue (VAT) (-21.6 cm2, P = .012). Intervention group II (low dose): Reduced waist circumferences significantly (P=0.045).

Table 2 Summary of RCTs about effect of probiotics on T2D.

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Tonucci et al 2017[22]	Brazil	Randomized, double-blind, placebo-controlled study, 6 weeks	Adults with BMI lower than 35 kg/m^2 and type 2 diabetes diagnosed for at least one year Age: 35-60 y Male: 26 (57.8%) Female: 19 (42.2%)	S. thermophilus TA-40, L. acidophilus La-5, lactis BB-12	Total: 45 Placebo group: n=22 Intervention group: n=23	Depends on the date	Placebo	Intervention group: reduced fructosamine levels (-9.91 mmol/L; p=0.04), hemoglobin A1c (-0.67%; p=0.06); HbA1c (-0.31 for control group vs -0.65 for probiotic group; p=0.02), total cholesterol (+0.55 for control group vs -0.15 for probiotic group; p=0.04) and LDL-cholesterol (+0.36 for control group vs -0.20 for probiotic group, p=0.03).
Sabico et al 2017[23]	the United Kingdom	Single-center, double- blind, randomized, placebo-controlled study, 3 months	Adult Saudis with newly diagnosed T2DM Age: 30–60 y Male: 40 (51.3%) Female: 38 (48.7%)	Sachets with 2 g freeze-dried powder of the probiotic mixture ®Ecologic®Barrier (Winclove9probiotics, the Netherlands). Ecologic Barrier (2.5×10^9 cfu/g) contains the following bacterial strains: Bifidobacterium bifidum W23, Bifi- dobacterium lactis W52, Lactobacillus acidophilus W37, Lactobacillus brevis W63, Lactobacillus casei W56, Lac- tobacillus salivarius W24, Lactococcus	Total: 78 Placebo group: n=39 Intervention group: n=39	5×10^9 cfu/day	Placebo	Intervention group: no difference in endotoxin levels between groups; significant improvement is shown in WHR and HOMA-IR.
Frouzi et al 2017[24]	Malaysia	Randomized, double-blind, parallel-group, con- trolled clinical trial, 12 weeks	T2DM patients with established T2D for at least 6 months prior to the commencement of the study, not on insulin and antibiotics, had glycated hemoglobin A1c (HbA1c) between 6.5 and 12 %, had fasting blood glucose (FBG) <15 mmol/L, had BMI between 18.5 and 40 kg/m2, and on a stabilized dose of medication for at least 3 months before the study. Age: 30–70 y	Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus lactis; Bifidobacte- rium bifidum, Bifidobacterium longum and Bifidobacterium infantis	Total: 136 Placebo group: n=68 Intervention group: n=68	1×10^9 cfu/day for each strain	Placebo	Intervention group: glycated hemoglobin decreased by 0.14 % PP analysis ($p < 0.05$, small effect size of 0.050), but not significant in ITT analysis. Fasting insulin decreased by 2.9 μU/mL in probiotics group in PP analysis, and was significant between groups at both analyses ($p < 0.05$, medium effect size of 0.062 in PP analysis and small effect size of 0.033 in ITT analysis)

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Table 2 (continued)

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Sabico et al 2018[25]	the United Kingdom	Randomized, double-blind, placebo-controlled trial, 6 months	Adult Saudis with newly diagnosed T2DM Age: 30–60 y Male: 40 (51.3%) Female: 38 (48.7%)	Sachets with 2 g freeze-dried powder of the probiotic mixture ®Ecologic®Barrier (Winclove9probiotics, the Netherlands). Ecologic Barrier (2.5×10^9 cfu/g) contains the following bacterial strains: Bifidobacterium bifidum W23, Bifi- dobacterium lactis W52, Lactobacillus acidophilus W37, Lactobacillus brevis W63, Lactobacillus casei W56, Lac- tobacillus salivarius W24, Lactococcus lactis W19 and Lactococcus lactis W58.	Total: 78 Placebo group: n=39 Intervention group: n=39	5×10^9 cfu/day	Placebo	Intervention group: significant decreases in circulating levels of endotoxin by almost 70% over 6 months, glucose (38%), insulin (38%), HOMA-IR (64%), triglycerides (48%), total cholesterol (19%), total/HDL-cholesterol ratio (19%), TNF-a (67%), IL-6 (77%), CRP (53%), resistin (53%), and a significant increase in adiponectin (72%) as compared with baseline. Only HOMA-IR had a clinically significant reduction (-3.4, 64.2%) in the intervention group group as compared to placebo group at all time points.
Raygan et al 2018[26]	Iran	Randomized, double-blind, placebo-con- trolled trial, 12 weeks	Patients with T2DM and 2- and 3-vessel CHD Age: 40–85 y	Bifidobacte- rium bifidum, Lactobacillus casei, Lacto- bacillus acidophilus	Total: 60 Placebo group: n=30 Intervention group: n=30	2×10^9 cfu/day for each strain	Placebo	Intervention group: significantly decreased fasting plasma glucose, insulin, insulin resistance and total-/HDL-cholesterol ratio, and significantly increased insulin sensitivity and HDL-cholesterol levels compared with the placebo.
Khalili et al 2018[27]	Iran	Parallel-group, randomized controlled trial, 8 weeks	Patients with T2DM Male: 14 (35%) Female: 26 (65%)	L. casei	Total: 40 Placebo group: n=20 Intervention group: n=20	10^8 cfu/day	Placebo	Intervention group: significantly decreased fasting blood sugar, insulin concentration, and insulin resistance compared with placebo group; insignificant decrease of HbA1c.

Table 3 Summary of RCTs about effect of probiotics on digestive health.

Study(year)	**Country**	**Study design and duration**	**Population, sex and age(y)**	**Strain of probiotcis**	**Sample size (n)**	**Probiotics dose**	**Control group**	**Results(Intervention vs control after supplementation)**
Tannaka et al 2015[28]	Japan	Double-blind, parallel-group comparison trial, 8 weeks	Female with mildly constipation Age: 25-49 y	Bifidobacterium animalis subsp. lactis GCL2505 (B. lactis GCL2505)	Total: 38 Placebo group: 20 Intervention group: 18	100g test beveage/day	Placebo	Intervention group: significantly increased number of fecal bifidobacteria and the frequency of defecation.
Mirghafourvand et al 2016[29]	Iran	Triple-blind randomized controlled trial, 4 weeks	Healthy singleton pregnant women who suffered from constipation	Bifidobacterium, Lactobacillus	Total: 60 Placebo group: 30 Intervention group: 30	4.8×10^{10} cfu/day	Placebo	Intervention group: significantly increased frequency of defecation; greatly improved constipation symptoms including straining, anorectal obstruction, manipulation to facilitate defecation, consistency of stool and color of stool.
Ding et al 2016[30]	China	Randomized, placebo-controolled trial, 12 weeks	Adults with BMI 18.5–25 kg/m^2 and disgnosed chronic constipation, complete bowel movements (SCBMs) per week for a minimum of 6 months, colonic transit time (CTT) >48 h, mild-to-moderate constipation with a Wexner constipation scale score between 16 and 25	synbiotic (BIFICOPEC) whih contained 0.63 g of bifid triple viable capsules (BIFICO)	Total: 93 Placebo group: 45 Intervention group: 48	two synbiotics capsules/day	Placebo	Intervention group: increased stool frequency, stool consistency, decreased CTT, and improved constipation-related symptoms.
Ibarra et al 2018[31]	Finland	3-arm parallel-group (alloca- tion ratio 1:1:1), double-blind, randomized, placebo-controlled, monocenter trial, 28 days	Patients with BMI 18.5 to 34.9 kg/m^2, diagnosed with functional constipation by the investigator per the Rome III criteria in the last 3 months, with symptom onset occurring at least 6 months prior to the diagnosis Age: 18-70 y	Bifidobacterium animalis subsp. lactis HN019 (HN019)	Total: 228 Placebo group: 76 Intervention group (low dose): 76 Intercention group (high dose): 76	Intervention group (low dose): 10^9 cfu/day Intercention group (high dose): 10^{10} cfu/day	Placebo	Intervention group: no statistically significant differences.

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Table 3 (continued)

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Escribano et al 2018[32]	Spain	Multicenter, double-blind, randomized, placebo-controlled parallel group trial, 12 weeks	Infants with normal birth weight (3rd to 97th percentiles for gestational age), normal growth curve according to current Spanish references, and ≤3 months of age at enrolment Age: ≤ 3 months Male: 69 (45.7%) Female: 82 (54.3%)	B. infantis IM1	Total: 151 Placebo group: 78 Intervention group: 73	10^7 cfu/day	Placebo	Intervention group: lower constipation incidence, higher stool cnosistency and less diarrhea episodes.
Yoon et al 2018[33]	Korea	Randomized, double-blind, placebo-controlled trial, 4 weeks	Patients with diagnosis of IBS-C or FC (based on Rome IV criteria) Age: 18-70 y Male: 24 (14.0%) Female: 147 (86.0%)	Streptococcus thermophilus MG510, Lactobacillus plantarum LRCC5193	Total: 171 Placebo group: 83 Intervention group: 88	3.0 × 108 cfu/day of Streptococcus thermophilus MG510 and 1.0 × 108 cfu/day of Lactobacillus plantarum LRCC5193	Placebo	Intervention group: significantly improved stool consistency measured by the Bristol Stool Form Scale (3.7 ± 1.1 vs. 3.1 ± 1.1 at 8 weeks, P = 0.002) and significantly bettter quality of life (at 8 weeks p = 0.049). No significant difference was found in CSBM.
Gomi et al 2018[34]	Japan	Double-blind, randomized, placebo-controlled trial, 4 weeks	Adults with temporary gastric symptoms with a modified Frequency Scale for Symptoms of Gastroesopha-geal reflux disease (m-FSSG) score ≥8 but were not defined as having " functional dyspepsia" by the Rome IV classification Age: 20-64 y Male: 38 (48.1%) Female: 41 (51.9%)	Bifidobacterium bifidum YIT 10347 (YIT10347)	Total: 79 Placebo group: 40 Intervention group: 39	Intervention group YIT10347- 100 ml fermented milk per day which contained more than 3 × 10^7 cfu/mL of YIT10347 and more than 1 × 10^7 cfu/mL of S. thermophilus YIT 2021. Placebo groupe: 100 ml placebo milk per day which contained more than 1 × 10^7 cfu/mL of S. thermophilus YIT 2021 only.	Placebo milk	Intervention group: higher relief rates of overall gastrointestinal symptoms, upper gastrointestinal symptoms, flatus, and diarrhea.

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Table 3 (continued)

Study(year)	Country	Study design and duration	Population, sex and age(y)	Strain of probiotcis	Sample size (n)	Probiotics dose	Control group	Results(Intervention vs control after supplementation)
Sun et al 2018[35]	China	Multicenter, randomized, double-blind, placebo-controlled trial, 4 weeks	Patients diagnosed with diarrhea-predominant IBS (IBS-D) Age: 18-65 y Male: 116 (58%) Female:84 (42%)	Clostridium butyricum	Total: 200 Placebo group: 95 Intervention group: 105	1.5×10^{7}/day	Placebo	Intervention group: improved overall IBS-D symptoms (−62.12 ± 74.00 vs. −40.74 ± 63.67, p=0.038) and quality of life (7.232 ± 14.06 vs. 3.159 ± 11.73, p=0.032) and stool frequency (−1.602 ± 1.416 vs. −1.086 ± 1.644, p=0.035).
Shin et al 2018[36]	Korea	Single center, randomized, double-blind and placebo-controlled clinical trial, 8 weeks	Subjects with diarrhea dominant IBS according to the Rome III criteria Age: 20–55 y Male: 22 (43.1%) Female:29 (56.9%)	Lactobacillus gasseri BNR17	Total: 51 Placebo group: 27 Intervention group: 24	10^{10} cfu/day	Placebo	Intervention group: significantly improvement of the symptoms of diarrhea, abdominal pain, distension, disturbed daily life, and mean defecation frequency.

CONCLUSIONS

Randomized clinical trials in recent three years have statistically suggested that intake of probiotics have a beneficial impact on obesity and T2DM, but not significant enough on digestive health. Considering the complexity of probiotics strains and consistency of data collected from these trials, more RCTs are necessarily needed to clarify the effect of probiotics in detail.

Up to date, a great number of experiments or clinical trials on the effect of probiotics have provided evidence that probiotics can influence the composition of gut microbiota, and bring positive effect on human health. It is a promising therapeutic treatment for us. However, due to the complicated human body structure and difference among people's dietary routine, genetic background, and many other factors, probiotics may not have the same effect on everybody, and not all species of probiotics are good for us. All scientific and medical researchers agree that there is not enough evidence to reliably assess the possible role of probiotics in the treatment of all health disorders. In order to get more reliable results, further studies with a higher number of patients and animals should be done. Also, the specific assessment of probiotics function should also be clarified as a standard in all experiments.

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